

Application No. 10/597,636

REQUEST FOR CONTINUED EXAMINATION AND AMENDMENT dated July 6, 2011

Reply to Office Action of May 11, 2011

Attorney Docket 9248-88834-US

AMENDMENTS TO THE CLAIMS

This listing of claims will replace all prior versions, and listings, of claims in the application:

Listing of Claims:

1. – 14. (Canceled)

15. (Currently Amended) A method for measuring the activation status of neutrophil cells in a biological sample obtained from a mammal, the sample containing the neutrophil cells and/or enzyme released by the neutrophil cells, which method specifically measures the active enzyme content only, the content being correlated with the neutrophil cell activation status, the method comprising the steps of:

- immunocapturing the enzyme released by the neutrophil cells present in the biological sample by contacting the biological sample with an enzyme specific polyclonal or monoclonal antibody; and
- detecting and/or measuring ~~active~~ the activity of the immunocaptured enzyme present ~~in said biological sample~~ which indicates the activation status of neutrophil cells in the biological sample.

16. (Canceled)

17. (Previously Presented) The method according to claim 15 which is a Specific Immunological Extraction Followed by Enzymatic Detection method (SIEFED), wherein the step of capturing the enzyme is followed by washing to remove any components that can interfere with the measurement of the enzyme activity, the enzymatic activity of the enzyme

fixed by its specific antibody being detected by adding a specific substrate to be transformed by the active enzyme into a detectable reaction product.

18. (Previously Presented) The method according to claim 17, wherein H₂O₂ is added to the reaction medium and the substrate is a fluorimetric reaction product.

19. (Currently Amended) The method according to claim 15, wherein the biological sample is a cellular or acellular sample selected from the group consisting of arterial, venous and capillary blood, serum, plasma, seminal fluid, broncho-alveolar fluid, urine, saliva, endotracheal fluid, peritoneal fluid, uterine irrigation liquids, sputum, synovial fluid, ~~broncho-alveolar fluid~~, nasal fluid, gastric bowel and faecal derivate samples, cerebrospinal fluid and tissue extracts.

20. (Previously Presented) The method according to claim 15, wherein the neutrophil cell activation status is measured and correlated to a disease and/or pathology.

21. (Currently Amended) The method of claim 15, which further comprises the steps of:

comparing the active enzyme values from subjects known to present an activation of neutrophils with normal enzyme levels obtained from subjects without diseases ~~known to present an activation of neutrophils~~; and

relating the active enzyme levels measured to a neutrophil cell activation status indicative of the presence or absence of inflammatory diseases or immunological diseases affecting the neutrophil activation status in mammals.

22. (Previously Presented) The method of claim 15, wherein the mammal is a horse.

23. (Previously Presented) The method of claim 18, wherein an effective amount of nitrite is added to the reaction medium to enhance the generated fluorescent signal.

24. (Withdrawn) The method according to claim 15:

for the detection and/or the prediction of a disease or pathology selected from the group consisting of chronic or acute inflammatory diseases, digestive pathologies, strangulated intestinal pathologies, sepsis, septic shock, chronic and acute pulmonary pathologies, ischemia-reperfusion pathologies, articular pathologies, colics, laminitis, allergies, infections and cardiovascular diseases,

- to follow-up neutrophil cell activation during therapy of a diseased mammal,
- to evaluate the ability of neutrophil cells and/or drugs to fight against micro-organisms and/or to destroy them,
- to evaluate the efficiency of immunomodulators or the *in vitro* inhibitory capacity of drugs by comparing the neutrophil activation status of treated and non-treated neutrophils,
- to evaluate the ability of neutrophils treated with modulators and/or drugs to against micro-organisms and/or to destroy them,
- to evaluate the natural defense capacity or ability of a mammal to fight against micro-organisms,
- to screen and to select compounds which interact with myeloperoxidase (MPO) and possibly inhibit myeloperoxidase (MPO) activity or
- to distinguish between total and active myeloperoxidase (MPO) content in the biological sample.

25. (Canceled)

26. (Currently Amended) A Specific Immunological Extraction Followed by Enzymatic Detection (SIEFED) kit ~~or device~~ for measuring the activation status of neutrophil cells in a biological sample obtained from a mammal, which kit ~~or device~~ specifically measures the active enzyme content only, the content being correlated with the cell activation status, the kit ~~or device~~ comprising:

- antibodies effective for immunocapturing the enzyme released by the neutrophil cells present in a biological sample obtained from a mammal, ~~the sample containing the neutrophil cells or the enzyme released by the cells~~, and
- a substrate effective for detecting and/or measuring ~~active~~ the activity of the immunocaptured enzyme present which indicates the activation status of the neutrophil cells in the biological sample.

27. (Withdrawn) A screening and selection method of compound(s) interacting with myeloperoxidase (MPO), which comprises the steps of:

- capturing active myeloperoxidase (MPO) to a solid support,
- adding one or more compound(s) to the active myeloperoxidase (MPO),
- measuring myeloperoxidase (MPO) activity after addition of the compound(s).

28. (Withdrawn) The method of claim 27, wherein the step of capturing active myeloperoxidase (MPO) is done by an antibody.

29. (Withdrawn) The method according to the claim 27, which comprises after the step of adding one or more compound(s) to the active myeloperoxidase (MPO) a step of washing of the compound(s) which are not bound to the active myeloperoxidase (MPO).

30. (Withdrawn) The method of claim 27 for the screening selection of compounds inhibiting myeloperoxidase (MPO) activity.

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31. (Withdrawn) The method of claim 27 which further comprises the step of:
- measuring myeloperoxidase (MPO) activity before addition of the compounds,
 - comparing myeloperoxidase (MPO) activities before or after addition of the compounds,
- and
- recovering the compounds that interact with myeloperoxidase (MPO).

32. (Previously Presented) The method of claim 15, wherein the enzyme is myeloperoxidase.

33. (Canceled)

34. (Previously Presented) The method of claim 15, wherein the enzyme is elastase.

35. (Previously Presented) The method of claim 18, wherein the substrate is 10-acetyl-3, 7-dihydroxyphenoxazine.

36. (Previously Presented) The method of claim 21, which further comprises comparing and correlating the quantified active enzyme level with a standard active enzyme curve.

37. (Currently Amended) The kit ~~or device~~ of claim 26, wherein the enzyme is myeloperoxidase.

38. (Canceled)

39. (Currently Amended) The kit ~~or device~~ of claim 26, wherein the enzyme is elastase.

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40. (Previously Presented) The method of claim 15, further comprising the step of detecting and/or measuring the total active and inactive enzyme present in the biological sample by a second enzymatically labeled enzyme specific polyclonal or monoclonal antibody.